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3M

October 26, 2004

8EHQ-1004-15974S

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Office of Pollution, Prevention and Toxics
U.S. Environmental Protection Agency
1200 Pennsylvania Avenue, N. W.
Washington, DC 20460
Attention: Section 8(e) Coordinator

Re: **TSCA Section 8(e) Submissions**

Dear Sir/Madam:

3M Company ("3M") requests that EPA place the attached studies in the TSCA Section 8(e) docket. We have included a master index for these studies identifying the study title, test substance and CAS number. A Confidential Business Information (CBI) version of this index and the studies also is being submitted today pursuant to EPA procedures. 3M has not provided CBI substantiation with this submission, but would be willing to do so at the Agency's request.

3M has concluded that data in these studies may not be, strictly speaking, "corroborative" of previously reported or published information as defined in EPA's reporting guidance or otherwise potentially may warrant 8(e) submission based on EPA's reporting guidance.

3M appreciates EPA's attention to this matter. Please contact the undersigned if you have any questions or require further information regarding this submission.

Very truly yours,

Katherine E. Reed (9.7.)

Dr. Katherine E. Reed, Ph.D
Staff Vice President
Environmental Technology and Safety Services
(651) 778-4331
kereed@mmm.com

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Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
(Confidential Business Information Redacted)

Title	Substance Information	CAS Information
Aquatic Toxicity Data Sheet: 48hr *Daphnia Magna*	1,4-dioxane; heptadecafluoro-1-octanesulfonic acid; linear n-ethyl perfluorooctanesulfonamide; n-ethylperfluorooctanesulfonamidoethyl alcohol; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(heptadecafluorooctyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(nonafluorobutyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(pentadecafluoroheptyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(tridecafluorohexyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(undecafluoropentyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; polyethylene glycol; water	1,4-dioxane (123-91-1); heptadecafluoro-1-octanesulfonic acid (1763-23-1); linear n-ethyl perfluorooctanesulfonamide (4151-50-2); n-ethylperfluorooctanesulfonamidoethyl alcohol (1691-99-2); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(heptadecafluorooctyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (29117-08-6); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(nonafluorobutyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-79-3); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(pentadecafluoroheptyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-81-7); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(tridecafluorohexyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (56372-23-7); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(undecafluoropentyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-80-6); polyethylene glycol (25322-68-3); water (7732-18-5)
Multigeneration Daphnid Life Cycle Test	1,4-dioxane; heptadecafluoro-1-octanesulfonic acid; linear n-ethyl perfluorooctanesulfonamide; n-ethylperfluorooctanesulfonamidoethyl alcohol; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(heptadecafluorooctyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(nonafluorobutyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(pentadecafluoroheptyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(tridecafluorohexyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(undecafluoropentyl)sulfonyl]amino]ethyl]-.omega.-hydroxy-; polyethylene glycol; water	1,4-dioxane (123-91-1); heptadecafluoro-1-octanesulfonic acid (1763-23-1); linear n-ethyl perfluorooctanesulfonamide (4151-50-2); n-ethylperfluorooctanesulfonamidoethyl alcohol (1691-99-2); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(heptadecafluorooctyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (29117-08-6); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(nonafluorobutyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-79-3); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(pentadecafluoroheptyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-81-7); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(tridecafluorohexyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (56372-23-7); poly(oxy-1,2-ethanediyl), .alpha.-[2-[ethyl[(undecafluoropentyl)sulfonyl]amino]ethyl]-.omega.-hydroxy- (68298-80-6); polyethylene glycol (25322-68-3); water (7732-18-5)
Aquatic Invertebrate Testing - Alkyltins LR 8024-1	Alkyltins: dibutyltin laurate and dibutyltin-di(2 ethylhexoate)	Dibutyltin laurate (CAS 77-58-7); Dibutyltin-di(2 ethylhexoate) (CAS 2781-10-4)
Aquatic Invertebrate Testing - Decosheen Material (LR-8052)	Decosheen Ribbon Materials and pigments: Decosheen Blue in Green Ceres Blue ZV; Decosheen Gold Paste Pigment; Decosheen Royal Blue, Solvent Blue	Decosheen Blue in Green [redacted] Blue ZV (CAS 61814-09-3); Decosheen Royal Blue, Solvent Blue [redacted]-86-6); Decosheen Gold Paste Pigment (CAS Number [redacted]
R Scratch Remover (Fathead Minnow)	55-65% Water; 20-30% Stoddard Solvent; 1-5% Sodium Silicate; 1-5% Potassium Hydroxide; 0.1-3% Nonylphenoxypoly(oxyethylene)ethanol	Water (CAS 7732-18-5); Stoddard Solvent (CAS 8052-41-3); Sodium Silicate (CAS 1344-09-8); Potassium Hydroxide (CAS 1310-58-3); Nonylphenoxypoly(oxyethylene)ethanol (CAS 9016-45-9)
S Scratch Remover (Fathead Minnow)	60-70% Water; 20-30% Stoddard Solvent; 1-5% Sodium Silicate; 0.1-3% Turgitol NP-33	Water (CAS 7732-18-5); Stoddard Solvent (CAS 8052-41-3); Sodium Silicate (CAS 1344-09-8); Turgitol NP-33 (CAS 9016-45-9)
Octanol Water Partition Coefficient	N-methylperfluorooctane sulfonamidoethanol	CAS 24448-09-7

Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
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Title	Substance Information	CAS Information
CoCl2.6H2O as Co2+ Toxicity to Microtox Reagent	Cobalt (as Co2+ ion) (CoCl2.6H2O)	CAS 7791-13-1
Activated Sludge Respiration Inhibition Test on CoCl2.6H2O as Co ion	Cobalt (as Co2+ ion) (CoCl2.6H2O)	CAS 7791-13-1
Acute Toxicity of CoCl2.6H20 as Co ion to Daphnia magna under Static Exposure Conditions	Cobalt (as Co2+ ion) (CoCl2.6H2O)	CAS 7791-13-1
Acute Toxicity of CoCl2.6H20 as Co ion to Fathead Minnow under Static Exposure Conditions	Cobalt (as Co2+ ion) (CoCl2.6H2O)	CAS 7791-13-1
Freshwater Algae Growth Inhibition Test	Cobalt (as Co2+ ion) (CoCl2.6H2O)	CAS 7791-13-1
Daphnia magna 21-Day Chronic Reproduction Study	N-ethylperfluorooctane sulfonamidoethanol	CAS 1691-99-2
Plant Growth Effects of [ ]	[ ]	[ ]
Final Report (*Daphnia* and Microtox)	Monomethyl ether of hydroquinone	CAS 150-76-5
Microtox Test Results	2 Ethylhexyl Acrylate; Isooctyl Acrylate Monomer; 2-Methylbutyl acrylate; Methyl isoamyl acrylate; Isooctyl Acrylate	2 Ethylhexyl Acrylate (CAS 103-11-7); Isooctyl Acrylate Monomer (CAS 29590-42-9) 2-Methylbutyl acrylate (CAS 44914-03-6); Methyl isoamyl acrylate (CAS 18993-92-1); Isooctyl Acrylate (CAS 29590-42-9)
Phytotoxicity Test Results	[ ]	[]

Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
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Title	Substance Information	CAS Information
Plant Toxicity Comparison, Young Seedling Growth	[ ]	[ ]
Ceriodaphnia dubia Survival and Reproduction exposed to Opequon Creek Water Spiked with BETZ 1110 Polymer (November 4, 1987 sample) for seven days under static renewal conditions	BETZ 1110: Non-3M Product - Chemical composition not provided to 3M by manufacturer	MSDS provided by manufacturer states product is "not hazardous" and not "considered to be a carcinogen"
Ceriodaphnia dubia Survival and Reproduction exposed to Opequon Creek Water Spiked with Betz 1138 Polymer (November 4, 1987 sample) for seven days under static renewal conditions	BETZ 1138: Non-3M Product - Chemical composition not provided to 3M by manufacturer	MSDS provided by manufacturer states product is "not hazardous" and not "considered to be a carcinogen"
Toxicity of 1,6 - Hexanediol Diacrylate to *Daphnia magna*	1,6 Hexanediol diacrylate	CAS 13048-33-4
Daphnia magna Chronic Bioassay Under Static Renewal Conditions	Methyl isoamyl acrylate	CAS 18993-92-1
Estimating the Chronic Toxicity of Nalclear 7177 to *Ceriodaphnia* Survival and Reproduction Using Short-Term Tests	Nalclear 7177 wastewater treatment acrylamide/acrylate polymer - Chemical composition not provided to 3M by manufacturer	CAS Information not provided to 3M by manufacturer
Acute Toxicity of Isooctyl Acrylate to *Daphnia magna*	Isooctyl Acrylate Monomer	CAS 29590-42-9
Static Acute Toxicity of [ ] to the Daphnid, *Daphnia magna*	Tolyltriazole	CAS 29385-43-1
Static Acute Toxicity of [ ] to the Alga, *Selenastrum capricornutum*	Tolyltriazole	CAS 29385-43-1
Static Acute Toxicity of [ ] to the Daphnid, *Daphnia magna*	[ ]	[ ]
Static Acute Toxicity of [ ] to the Fathead Minnow, *Pimephales promelas*	[ ]	[ ]
Static Acute Toxicity of [ ] to the Daphnid, *Daphnia magna*	water; propylene-tetrafluoroethylene polymer; tert-butyl alcohol	water (7732-18-5); propylene-tetrafluoroethylene polymer (27029-05-6); tert-butyl alcohol (75-65-0)

Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
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Title	Substance Information	CAS Information
Isooctyl acrylate: Fish, Acute Toxicity Test	Isooctyl Acrylate Monomer	CAS 29590-42-9
Isooctyl Acrylate: *Daphnia sp.* Acute Immobilization Test	Isooctyl Acrylate Monomer	CAS 29590-42-9
Isooctyl Acrylate: Alga, Growth Inhibition Test	Isooctyl Acrylate Monomer	CAS 29590-42-9
Isooctyl Acrylate: *Daphnia sp.* Reproduction Test	Isooctyl Acrylate Monomer	CAS 29590-42-9
Acute Toxicity of [ ] to the mysid, *Mysidopsis bahia*	[ ]	[ ]
Final Report (Microtox)	[ ]	[ ]
Determination of the Partition Coefficient (N-Octanol/Water) of T-5896 by High Performance Liquid Chromatography (HPLC)	N-methyl perfluorooctane sulfonamido ethanol; N-methyl perfluorooctane sulfonamidethyl acrylate	N-methyl perfluorooctane sulfonamido ethanol (CAS 25268-77-3); N-methyl perfluorooctane sulfonamidethyl acrylate (CAS 24448-09-7)
OECD Activated Sludge Respiration Inhibition Test Results	N-Dodecyltrimethylammonium chloride	CAS = 112-00-5
Final Report (Fish Acute Toxicity)	Mirataine CB (30% Cocamidopropyl betaine = Amides, coco, N-(3-(dimethylamino)propyl), alkylation products with chloroacetic acid, sodium salts, 70% Water and Inerts); Mirataine COB (30% Coco/Oleamidopropyl Betaine = 1-Propanaminium, 3-amino-N-(carboxymethyl)-N,N-dimethyl-, N-coco acyl derivs., inner salt)	Cocamidopropyl betaine (CAS 70851-07-9); Coco/Oleamidopropyl Betaine (CAS 61789-40-0)
A Flow-Through Life-Cycle Toxicity Test With the Saltwater Mysid (*Mysidopsis bahia*)	Perfluorooctane sulfonate	CAS 1763-23-1
Lithium: Alga, Acute toxicity Tests	Lithium Chloride	CAS 7447-41-8
An Early Life-Stage Toxicity Test With the Fathead Minnow (*Pimephales promelas*)	Perfluorooctane sulfonate	CAS 1763-23-1
Lithium: Fish, Acute toxicity Tests	Lithium Chloride	CAS 7447-41-8
Lithium: *Daphnia*, Acute toxicity Tests	Lithium Chloride	CAS 7447-41-8
Summary of Toxicity Testing on OSCl and OSF	Octane sulfonyl chloride and Octane sulfonyl fluoride	Octane sulfonyl fluoride (CAS 7795-95-1), Octane sulfonyl chloride (CAS 4063-63-5)
Toxicity to Microtox Test	Lauryldimethylamineoxide	CAS 1643-20-5

Master Index to Studies Submitted Under TSCA 8(e) by 3M Company on October 26, 2004
(Confidential Business Information Redacted)

Title	Substance Information	CAS Information
Ecotoxicological Testing of CoCl2.6H2O as Co2+ ion (Seed Germination and Root Elongation)	Cobalt (as Co2+ ion) (CoCl2.6H2O)	CAS 7791-13-1

CONFIDENTIAL BUSINESS INFORMATION
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STUDY TITLE

ISOOCTYL ACRYLATE: FISH, ACUTE TOXICITY TEST

DATA STANDARD

OECD GUIDELINE 203

AUTHORS

Joe Amato and Dinesh Vaishnav

STUDY COMPLETED

May 28, 1992

TESTING FACILITY

AScI Corporation
AScI-Duluth Environmental Testing Division
112 East Second Street
Duluth, MN 55805

Tel. No. (218) 722-4040

STUDY IDENTIFICATION NUMBERS

AScI Study ID# 5030-003-09

3M Company Study ID# J2774

CERTIFICATION OF GOOD LABORATORY PRACTICE COMPLIANCE

To the best of my knowledge, this study was conducted in accordance with OECD Good Laboratory Practice Standards (OECD Council Decision C(81)30, Annex 2: OECD Principles of Good Laboratory Practice 1981).

Study Director: _____ Date: 5/29/97
Joe Amato
ASci Corporation/ASci-Duluth
Environmental Testing Division

Based on the signatures of the Study Director and the Quality Assurance Auditor, this study, to the best of our knowledge, was conducted in accordance with OECD Good Laboratory Practice Standards (OECD Council Decision C(81)30, Annex 2: OECD Principles of Good Laboratory Practice 1981).

Sponsor: _____ Date: 5-29-97
Submitter: _____ Date: 5/29/97

STATEMENT OF QUALITY ASSURANCE

The study data were reviewed by the ASci-Duluth Environmental Testing Division Quality Assurance Unit to assure that standard operating procedures and guidelines used to conduct this study were followed, and this report is an accurate reflection of the raw data. The types of audits performed are listed in the following table.

Type of Audit for ASci Study ID# 5030-003-09	Audit Date	Date Reported to Study Director and Management
Study Plan	01-16-1992	01-16-1992
In-Life Phase	02-27-1992	02-27-1992
Raw Data and Draft Report	04-07-1992	04-08-1992
Final Report	05-28-1992	05-28-1992

Alan Mozol
Acting Manager, Quality Assurance Unit

Date: 5/28/92

TABLE OF CONTENTS

CONFIDENTIAL BUSINESS INFORMATION
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REDACTED FROM THIS PAGE

STUDY SUMMARY TABLE

Study Title	Isooctyl Acrylate: Fish, Acute Toxicity Test
Data Standard	OECD Guideline 203 (OECD 1984), and Good Laboratory Practice standards as promulgated under the OECD Council Decision C(81)30, Annex 2: OECD Principles of Good Laboratory Practice (OECD 1981).
Sponsor	Rich Purdy, 3M Environmental Laboratory, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55106; Tel No. (612) 778-5379.
Sponsor's Representative	Susan A. Beach, 3M Environmental Laboratory, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55106; Tel No. (612) 778-7452.
Testing Facility	ASci Corporation/ASci-Duluth Environmental Testing Division, 112 East Second Street, Duluth, MN 55805; Tel. No. (218) 722-4040.
Study Director	Joe Amato
Acting QAU Manager	Alan Mozol
Testing Facility Director	Donald Mount
Study Initiation Date	January 31, 1992
Test Dates	February 28-March 3, 1992
Test Substance	Isooctyl acrylate (CAS No. 29590-42-9,[ ]Lot 3290), 99.7% acrylate (as determined by Sponsor[ ]liquid.
Test Organism	Juvenile fathead minnows (*Pimephales promelas*), mean length 1.6 cm.

Test Description	(1) Test and control exposures (each 1.5 L) were established using a proportional diluter, (2) all exposures were incubated for 96 h, and exposure water hardness, alkalinity, conductivity, temperature, pH and DO were determined at appropriate time intervals, and test substance concentrations were measured daily, (3) test organisms were observed for mortality (effect), and (4) effect data were used to calculate LC50 and NOEC values
Test Results	Based on mean measured concentration, isooctyl acrylate 96-h LC50 and 96-h NOEC for fathead minnows (*P. promelas*) were 0.67 mg/L and 0.34 mg/L.
Location of Raw Data and Final Report	ASci Corporation/ASci-Duluth Environmental Testing Division, 112 East Second Street, Duluth, MN 55805; Tel. No. (218) 722-4040.

1.0 INTRODUCTION

The test substance, isooctyl acrylate, is an ester primarily made from isooctanol and acrylic acid. It has negligible solubility in freshwater and its acute toxicity to fish species is not known. The purpose of the present study was to determine if possible, the 96-h LC50 and 96-h NOEC (no observed effect concentration) of the test substance for fathead minnow (*Pimephales promelas*) under flow-through test conditions. The study was conducted according to the AScI study plan.

2.0 TEST METHODS

2.1 Test Substance. The test substance, isooctyl acrylate (CAS No. 29590-42-9, [ ] Lot 3290), was received at AScI on October 3, 1991 in one amber glass bottle placed in a sealed metal container. The test substance was stored at room temperature as received. According to a material safety data sheet and a written communication provided by the Sponsor, (1) the test substance was a clear, colorless, mobile liquid with acrylate odor, (2) the test substance has negligible water solubility and 1 mm Hg vapor pressure at 50°C, (3) the test substance is 99.75% acrylate as determined by Sponsor [ ] (4) the test substance is stable

and its biodegradation ranged from 59%-85% in five days, and (5) the test substance concentration in deionized water can be analyzed by a GC method. The Sponsor also has information that, based on the chemical structure, there will be essentially no dissociation of the test substance at environmental pH levels. The Sponsor suspects the test substance may have glass surface activity.

2.2 Test Substance Solutions. The test substance stock solution was prepared daily as follows:

(1) Added 260 µl (229 mg) of test substance to 4 L of dilution water contained in a 4-L glass bottle;

(2) Mechanically stirred the mixture at an ambient temperature (15-20°C) for 15 minutes and then sonicated it for 15 minutes;

(3) Stopped sonication, transferred mixture to a 19-L glass stock bottle containing 14 L of dilution water (total volume 18 L) and mixed the contents (the stock bottle was pre-conditioned for 10-12 h with 18 L of 12 mg/L test substance solution prepared in deionized water);

(4) Fitted a glass tube into the stock bottle so that one end of the tube remained just above the bottom of the stock bottle; and

(5) Connected the exposed end of the glass tube to a pre-calibrated metering pump to deliver the test substance stock solution to the test system (proportional diluter).

For use in this test, two replicates each of dilution water control and five test substance nominal concentrations were established (total 10 test + 2 control exposure chambers). All nominal concentrations were calculated based on the measured test substance stock solutions, calibration of the metering system and a dilution factor of 1.8. The mean test substance nominal concentrations were between 0.35 mg/L and 3.45 mg/L.

2.3 Test organisms. Test organisms were juvenile fathead minnows with a mean length of 1.6 ± 0.57 cm (based on lengths of fish from two control exposures). Fish were obtained from Aquatic Biosystems, Inc., Fort Collins, Colorado. They appeared healthy and 0.6% mortality occurred during the holding and acclimation period. The fish were held and acclimated for 14 days, during which they were exposed to dilution water for the last nine days.

2.4 Dilution water. Dilution water was shallow well water collected from the Two Harbors, (Minnesota) area. At test time, the water had

a hardness of 186-187 mg/L (as $CaCO_3$) and a pH of 8.12-8.25. The water was aerated for at least 24 h prior to use in the test.

The well water is analyzed annually and the most recent chemical analysis is provided in Appendix A.

2.5 Exposure chambers. Exposure chambers were 3-L rectangular glass tanks (each measuring about 19.5 cm length X 9.5 cm width X 15.5 cm height). During the test, the aqueous phase in these chambers was maintained at 1.5 L with exposure water. The chambers were kept covered, except when water chemistry determinations were made, samples of exposure water were collected for the test substance analysis or dead organisms were removed.

2.6 Test System. The test was conducted using a proportional diluter. Prior to test initiation, the diluter was calibrated and operated for 24 h to allow the test substance to reach a steady state in the exposure chambers. The flow rate was adjusted to provide 500 ml of exposure water per hour, that is, eight daily volume additions to individual exposure chambers. The flow splits were reassessed at test termination. The agreement between the initial and the final flow splits ranged from 97.7%-102.8%. The

daily utilization of the test substance stock solution ranged from 17.5-17.75 L.

2.7 Test Performance. To begin the test, 10 test organisms per exposure chamber were impartially distributed. The mean loading rate, based on fish biomass in the control exposure, was 0.02 g fish L^{-1} day^{-1}. The test organisms were fed up to 21 h before test initiation. During the test, water temperature was maintained at 20.4-21.2°C and daily photoperiod was maintained, using cool white fluorescent lamps, for 16 h light and 8 h dark periods.

Test organisms were observed for signs of stress and mortality at 3 h and 6 h after the test initiation, and then observed daily. Each time observations were made, all dead test organisms were removed from the exposure chambers.

2.8 Determination of Water Chemistry Parameters. During the test, (1) water chemistry parameters of total hardness and alkalinity were determined at test initiation, (2) pH, dissolved oxygen concentration (DO) and temperature were monitored daily, and (3) specific conductivity was recorded at test initiation and termination.

2.9 Test Substance Analysis. The test substance concentrations in individual and composite samples were analyzed according to the following schedule:

Type of Sample	Frequency of Sampling	Total Number of Samples Analyzed
New stock solution	0 h, 24 h, 48 h, 72 h and 96 h	10 samples (duplicate analysis/sampling period)
Test and control	0 h	6 composite samples
Test and control	24 h	6 composite samples
Test and control	48 h	5 composite and 2 individual samples
Test and control	72 h	5 composite samples
Test and control	96 h	10 individual samples

Samples were collected each day 2-4 h after the stock solution was renewed. The composite samples were prepared by combining 200 ml of replicate samples in 500-ml brown glass bottles. Each replicate sample was collected in order of low to high test substance concentration using a 250-ml brown glass bottle. A separate bottle was used for the control exposure. The same bottles were used throughout the test. The samples were extracted and analyzed using procedures described in the analytical method validation report (Appendix B). If required, the sample concentration was performed under a nitrogen stream before analyzing for the test substance.

2.10 Data Analysis. For LC50 calculations, the test organism mortality data and the test substance mean measured concentrations were analyzed using the trimmed Spearman-Karber method (Hamilton et. al. 1977), and for NOEC calculation, the data were analyzed using TOXSTAT (University of Wyoming 1989) statistical software. Each measured test substance concentration was corrected, as described later, for the well water spike recovery before calculating the mean measured concentration.

3.0 RESULTS

The mortality in 96 h ranged from 0% in control exposure and the lowest two test exposures to 100% in the highest test exposure (Table 1). Based on mean measured concentration, the test substance LC50 values for the test organisms ranged from 0.67 mg/L (96-h and 72-h LC50) to 0.86 mg/L (48-h LC50) and the 96-h NOEC was 0.34 mg/L (Table 2). The 24-h LC50 was not calculable because of insufficient mortality.

The data for standard (deionized water) and test (well water) matrix spike recoveries are presented in Table 3. The mean spike recovery from deionized water was 88 ± 15.3% and from well water 81 ± 15.4% (Table 3).

The test substance nominal concentrations are presented in Table 4. All nominal concentrations were calculated based on the measured test substance stock solutions (Table 4), calibration of the metering system and a dilution factor of 1.8. The mean nominal concentrations were: 0.35, 0.62, 1.12, 2.01 and 3.45 mg/L (Table 4).

The test substance measured concentrations and recoveries are presented in Table 5. The test substance daily measured concentrations were corrected for well water spike recovery for that particular day before calculating test substance recoveries from exposure water. For example, the test substance final measured concentrations were corrected for 77% recovery (Table 5), as well water spike recovery for 96 h was 77% (Table 3).

The test substance mean recoveries were between 24% and 51% with 0.62 mg/L and 3.45 mg/L test substance nominal concentrations, respectively (Table 5). Relatively poor recoveries (less than 70% of nominal concentrations), may be due to abiotic processes (ASTM 1989), such as loss of test substance due to adsorption, volatilization, etc. The daily analysis of the samples of exposure water did not show the presence of test substance degradates at any

noticeable level. Therefore, it is unlikely that biotic processes caused any loss of test substance. The test substance recovery data imply the reported LC50 values may be reproducible under the test conditions employed in this study.

At test initiation, the hardness of test solutions from 186-187 mg/L and alkalinity from 179-181 mg/L (both as $CaCO_3$) (Table 6). During the test, the test temperatures were between 20.4°C and 21.2°C (Table 7), DO concentrations were between 8.4 mg/L and 8.7 mg/L (Table 8), and pH were between 8.14 and 8.25 (Table 9). The initial and final conductivities ranged from 351-375 µmhos/cm (Table 10). All these values for the water chemistry parameters are within the acceptable limits for this test (ASTM 1980).

At test termination, the lengths and weights of fish from control exposures were determined. The mean length was 1.6 ± 0.57 cm and weight was 0.025 ± 0.012 g (Table 11). The fish loading rate was calculated based on the mean weight.

From the quality assurance standpoint, this test is acceptable because it complies with all acceptance criteria (Table 12).

4.0 CONCLUSIONS

Based on mean measured concentration, isooctyl acrylate 96-h LC50 and 96-h NOEC for the juvenile fathead minnows (*P. promelas*), as determined from the acute toxicity test, were 0.67 mg/L and 0.34 mg/L, respectively.

5.0 DEVIATIONS FROM APPROVED ASCI STUDY PLAN

The deviations which occurred while conducting this study were:

(1) To control the test substance degradation, the stock solution was prepared daily. To prepare the solution rapidly in order to keep-up with the daily stock utilization, the mixture of test substance and dilution water had to be vigorously stirred and sonicated.

(2) Accidently, the test organisms were fed at 21 h before test initiation.

(3) The schedule for analyzing test substance concentration was changed so that exposures with 100% dead organisms could be analyzed on the day of observation.

To the best of our current scientific knowledge and understanding, these deviations should have no effect on the results presented in this report.

6.0 REPORT SIGNATURE

Study Director: _____ Date: 5/28/93
Joe Amato
ASci Corporation/ASci-Duluth
Environmental Testing Division

7.0 REFERENCES

American Society for Testing and Materials (ASTM). 1980. Standard Practice for Conducting Acute Toxicity Tests With Fishes, Macroinvertebrates, and Amphibians. ASTM. Philadelphia, PA. Publication E 729-80.

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Organization for Economic Cooperation and Development (OECD). 1981/1984 OECD Guidelines for Testing of Chemicals. OECD Publication Information Center, Washington, DC.

University of Wyoming. 1989. Toxstat Version 3.1. Fish Physiology and Toxicology Laboratory, Department of Zoology and Physiology. University of Wyoming, Laramie, WY.

8.0 PERSONNEL INVOLVED IN STUDY AND THEIR RESPONSIBILITIES

Personnel	Responsibility
Joe Amato	Study Director
Minren Xu	Analytical chemistry
Billie Samson	Laboratory assistance
Linda Christensen	Laboratory assistance
Joe Dierkes	Holding/acclimating test organisms
Dave Nessa	Holding/acclimating test organisms
Romesh Lakhan	Glassware preparation
Dinesh Vaishnav	Report preparation
Alan Mozol	QA
Nancy Jordan	Archivist

Table 1. Isooctyl acrylate (test substance): Mortalities of fathead minnows (*P. promelas*)

Test substance mean measured concn (mg/L)[a]	Cumulative number of dead organisms and % mortality[b]					
	3 h	6 h	24 h	48 h	72 h	96 h
<MDL[c] (control)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
0.09	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
0.15	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)
0.34	0 (0)	0 (0)	0 (0)	1 (5)	1 (5)	1 (5)
0.82	0 (0)	0 (0)	0 (0)	7 (35)	13 (65)	13 (65)
1.75	0 (0)	0 (0)	1 (5)	20 (100)	20 (100)	20 (100)

[a]Each concentration was corrected for daily well water spike recovery.

[b]For each concentration, two replicate exposures were made with a total of 20 test organisms, and percentage mortality is given in parenthesis.

[c]Method detection limit (MDL) was 0.04 mg/L test substance.

Table 3. Isooctyl acrylate (test substance): Spike recoveries

Matrix	Time of Analysis	Test substance concn (mg/L)		% Recovery[a]
		Target	Measured	
Matrix-- Deionized water				
Method blank	0 h	0.0	<MDL[b]	
	24 h	0.0	<MDL	NC[c]
	48 h	0.0	<MDL	NC
	72 h	0.0	<MDL	NC
	96 h	0.0	<MDL	NC
Spike solution	0 h	0.22	0.24	NC
	24 h	0.22	0.15	109
	48 h	0.22	0.16	71
	72 h	0.22	0.21	75
	96 h	0.22	0.19	94
Mean spike recovery 88 ± 15.3%				89

Continued on the next page.

Table 3 (continued)

Matrix	Time of Analysis	Test substance concn (mg/L)		% Recovery[a]
		Target	Measured	
Matrix-- Well water				
Method blank[d]	0 h	0.0	<MDL	NC
	24 h	0.0	<MDL	NC
	48 h	0.0	<MDL	NC
	72 h	0.0	<MDL	NC
	96 h	0.0	<MDL	NC
Spike solution	0 h	0.22	0.23	104
	24 h	0.22	0.14	63
	48 h	0.22	0.16	74
	72 h	0.22	0.19	86
	96 h	0.22	0.17	77
Mean spike recovery 81 ± 15.4%				

[a]Some of the spike recoveries may include a small percentage of round-off error.

[b]Method detection limit (MDL) was 0.04 mg/L test substance.

[c]NC = not calculated.

[d]Data were from control exposure.

Table 4. Isooctyl acrylate (test substance): Stock solutions and nominal concentrations

Exposure No.	Rep	Exposure period and nominal exposure concentrations (mg/L)*					Mean ± SD (mg/L)
		0 h	24 h	48 h	72 h	96 h	
Stock solution	A	9.45	10.88	10.01	9.24	12.60	
	B	9.96	10.34	10.97	9.33	12.58	10.54 ± 1.234
1 (control)	A	0.0	0.0	0.0	0.0	0.0	
	B					0.0	0.0 ± 0.0
2	A	0.31	0.34	0.33	0.29	0.40	
	B					0.40	0.35 ± 0.046
3	A	0.55	0.61	0.60	0.53	0.72	
	B					0.72	0.62 ± 0.082
4	A	1.00	1.09	1.08	0.96	1.29	
	B					1.29	1.12 ± 0.142
5	A	1.80	1.96	1.94	1.72	2.33	
	B					2.33	2.01 ± 0.261
6 (highest concn)	A	3.24	3.54	3.50			
	B			3.50			3.45 ± 0.138

*All nominal concentrations were calculated based on measured test substance stock solutions, calibration of the metering system and a dilution factor of 1.8, and duplicate values were used to correspond with the duplicate test substance analysis.

Table 5. Isooctyl acrylate (test substance): Nominal and measured concentrations

Test substance mean nominal concn (mg/L)	Rep	Test substance measured concn (mg/L)[a]					Mean ± SD	% Recovery[b]
		0 h	24 h	48 h	72 h	96 h		
0.0 (control)	A	-[c]	-	-	-	<MDL[d]		
	B	-	-	-	-	<MDL		
	A11[e]	<MDL	<MDL	<MDL	<MDL	-	NC[f]	NC
0.35	A	-	-	-	-	0.09		
	B	-	-	-	-	0.09		
	A11	0.09	0.09	0.10	0.07	-	0.09 ± 0.008	26
0.62	A	-	-	-	-	0.14		
	B	-	-	-	-	0.14		
	A11	0.15	0.17	0.17	0.11	-	0.15 ± 0.024	24
1.12	A	-	-	-	-	0.35		
	B	-	-	-	-	0.34		
	A11	0.34	0.37	0.39	0.23	-	0.34 ± 0.056	30

Continued on the next page.

Table 5 (continued)

Test substance mean nominal concn (mg/L)	Rep	Test substance measured concn (mg/L)[a]					Mean ± SD	% Recovery[b]
		0 h	24 h	48 h	72 h	96 h		
2.01	A	-	-	-	-	0.85		
	B	-	-	-	-	0.76		
	All	0.99	0.92	0.89	0.50	-	0.82 ± 0.174	41
3.45	A	-	-	1.81	-	-		
	B	-	-	1.76	-	-		
	All	1.71	1.71	-	-	-	1.75 ± 0.047	51

[a]All measured concentrations were corrected for daily well water spike recovery.

[b]Percentage recovery = (mean measured concentration/nominal concentration) X 100.

[c]- = Not determined.

[d]Method detection limit (MDL) was 0.04 mg/L test substance.

[e]All = composite sample prepared from replicate samples.

[f]NC = not calculated.

Table 6. Isooctyl acrylate (test substance): Exposure water hardness and alkalinity (both as $CaCO_3$, mg/L) at test initiation

Test substance mean nominal concn (mg/L)	Hardness (acceptable range: 160-200)	Alkalinity
0.0 (control)	186	181
3.45	187	179

Table 7. Isooctyl acrylate (test substance): Exposure water temperatures (°C)

Test substance mean nominal concn (mg/L)	Rep	Time (h)					Range (acceptable range: 20-22°C)
		0 h	24 h	48 h	72 h	96 h	
0.0 (control)	A	20.6	21.2	20.6	20.8	20.8	
	B	20.6	20.9	20.8	20.9	20.7	20.6-21.2
0.35	A	20.4	20.9	20.8	20.8	20.8	
	B	20.5	20.8	21.0	20.9	20.8	20.4-21.0
0.62	A	20.4	20.9	20.8	21.0	20.8	
	B	20.4	20.9	20.7	20.9	20.8	20.4-21.0
1.12	A	20.4	20.8	21.0	21.0	20.8	
	B	20.5	20.8	20.9	20.9	20.8	20.4-21.0
2.01	A	20.4	20.8	20.8	20.8	20.9	
	B	20.4	20.8	20.9	20.9	20.7	20.4-20.9
3.45	A	20.4	20.8	20.8	-	-	
	B	20.4	20.8	20.7	-	-	20.4-20.8

*- = Not determined.

Table 8. Isooctyl acrylate (test substance): Exposure water DO (mg/L)

Test substance mean nominal concn (mg/L)	Rep	Time (h)					Range (minimum acceptable value: 5.5 mg/L)
		0 h	24 h	48 h	72 h	96 h	
0.0 (control)	A	8.7	8.5	8.6	8.6	8.7	
	B	8.7	8.4	8.5	8.6	8.6	8.4-8.7
0.35	A	8.7	8.5	8.6	8.6	8.6	
	B	8.7	8.5	8.6	8.5	8.6	8.5-8.7
0.62	A	8.7	8.4	8.6	8.6	8.6	
	B	8.6	8.4	8.6	8.5	8.6	8.4-8.7
1.12	A	8.7	8.5	8.7	8.6	8.5	
	B	8.6	8.5	8.7	8.6	8.6	8.5-8.7
2.01	A	8.6	8.5	8.7	8.6	8.6	
	B	8.7	8.5	8.8	8.6	8.6	8.5-8.7
3.45	A	8.7	8.6	8.7	-	-	
	B	8.7	8.5	8.7	-	-	8.5-8.7

'- = Not determined.

Table 9. Isooctyl acrylate (test substance): Exposure water pH

Test substance mean nominal concn (mg/L)	Rep	Time (h)					Range (acceptable range: 7.5-8.5)
		0 h	24 h	48 h	72 h	96 h	
0.0 (control)	A	8.15	8.16	8.22	8.22	8.19	
	B	8.16	8.15	8.21	8.23	8.25	8.15-8.25
0.35	A	8.15	8.14	8.22	8.23	8.22	
	B	8.18	8.17	8.22	8.21	8.22	8.14-8.23
0.62	A	8.19	8.15	8.22	8.24	8.21	
	B	8.18	8.15	8.23	8.23	8.22	8.15-8.24
1.12	A	8.18	8.20	8.21	8.23	8.23	
	B	8.18	8.15	8.22	8.23	8.23	8.15-8.23
2.01	A	8.16	8.15	8.21	8.24	8.23	
	B	8.16	8.15	8.20	8.24	8.16	8.15-8.24
3.45	A	8.14	8.12	8.22	-*	-	
	B	8.14	8.12	8.23	-	-	8.12-8.23

*- = Not determined.

Table 10. Isooctyl acrylate (test substance): Exposure water conductivity (µmhos/cm) at test initiation and termination

Test substance mean nominal concn (mg/L)	Rep	Time (h)		Range
		0 h	96 h	
0.0 (control)	A	362	358	
	B	364	351	351-364
0.35	A	363	360	
	B	360	359	359-363
0.62	A	362	361	
	B	363	360	360-363
1.12	A	360	364	
	B	359	361	359-364
2.01	A	365	365	
	B	361	375	361-375
3.45	A	361	363 (48 h)	
	B	360	367 (48 h)	360-367

Table 11. Isooctyl acrylate (test substance): Lengths and weights of fish from control exposures

Fish length (mm)		Fish weight (g)	
Control rep A	Control rep B	Control rep A	Control rep B
17	19	0.035	0.033
18	19	0.031	0.048
14	16	0.033	0.027
17	18	0.036	0.017
18	16	0.013	0.048
14	14	0.020	0.024
14	15	0.013	0.028
13	16	0.016	0.011
12	13	0.009	0.028
12	15	0.006	0.018
Mean ± SD (cm)	1.6 ± 0.57	Mean ± SD (g)	0.025 ± 0.012

Table 12. Isooctyl acrylate (test substance): QA criteria and test acceptability

Criterion	Results
Less than 10% of test organisms in dilution water control must be affected	0% affected
During the test, DO concentration must be maintained at a minimum of 60% of the air saturation value at the test temperature	The lowest DO concentration measured was 93% of air saturation value
Measured test substance concentrations must be less than 130% of the nominal concentrations	Measured concentrations were less than 130% of the nominal
Test duration must be 96 h	Test duration was 96 h

Appendix A

Chemical Analysis of Well Water

Chemical Analysis of Well Water[a]

Parameter	µg/L	MDL[b] (µg/L)
Aldrin	ND[c]	0.3
A-BHC	ND	3.0
B-BHC	ND	0.4
D-BHC	ND	4.0
Chlordane (Gamma)	ND	1.0
Chlordane (Alpha)	ND	1.0
4,4'DDD	ND	0.3
4,4'DDE	ND	0.3
4,4'DDT	ND	0.3
Dieldrin	ND	0.3
Endosulfan I	ND	1.0
Endosulfan II	ND	1.0
Endosulfan Sulfate	ND	1.0
Endrin	ND	1.0
Endrin Aldehyde	ND	0.2
Heptachlor	ND	0.05
Heptachlor Epoxide	ND	0.3
Lindane (G-BHC)	ND	0.1
Toxaphene	ND	2.0
Methoxychlor	ND	1.0
Endrin Ketone	ND	1.0
PCB 1016	ND	1.0
PCB 1221	ND	1.0
PCB 1232	ND	1.0
PCB 1242	ND	1.0
PCB 1248	ND	1.0
PCB 1254	ND	1.0
PCB 1260	ND	1.0

Parameter	µg/L	MDL[b] (µg/L)
Malod	ND	7.5
Demeton	ND	1.0
Ronnel	ND	0.5
Chlorpyrifos	ND	0.5
DEF	ND	0.5
Bolstar	ND	0.5
Phosalone	ND	0.5
Guthion	ND	5.0
Coumaphos	ND	5.0
Dichlorvos	ND	1.0
Mevinphos	ND	3.5
Trifluralin	ND	0.5
Ethoprop	ND	0.5
Phorate	ND	0.5
Dimethoate	ND	0.5
Methyl Parathion	ND	0.5
Merphos	ND	0.5
Fenthion	ND	0.5
Diphenamid	ND	0.5
Ethion	ND	0.5
Fenamiphos	ND	1.0
Carbophene	ND	1.0
Diazinon	ND	0.5
Dimethoate	ND	0.5
Malathion	ND	2.0
Parathion	ND	0.5
Methyl Trithion	ND	1.0
Prometone	ND	0.5
Trichloronat	ND	0.5

Parameter	Unit	Conc.
Total Suspended Solids	mg/L	< 4
Ammonia Nitrogen	mg/L	< 0.05
Total Kjeldahl Nitrogen	mg/L	0.18
Chemical Oxygen Demand	mg/L	9
Total Cyanide	mg/L	< 0.01
Aluminum	µg/L	< 100
Arsenic	µg/L	< 2
Cadmium	µg/L	< 0.5
Calcium	mg/L	46.3
Cobalt	µg/L	< 2
Chromium	µg/L	< 3
Copper	µg/L	13
Iron	µg/L	3
Lead	µg/L	< 2
Magnesium	mg/L	16.3
Mercury	µg/L	< 0.2
Nickel	µg/L	< 2
Potassium	mg/L	< 0.5
Selenium	µg/L	< 3
Silver	µg/L	< 1
Sodium	mg/L	6.5
Zinc	µg/L	30
Note: Ca/Mg = 2.83 and Na/K = > 13		

[a] The Water Was Collected from Two Hardrock Geothermal Areas and Analyzed During August-September 1991

[b] MDL = Method Detection Limit

[c] ND = Not Detected

Appendix B*

Isooctyl Acrylate: Method Validation for Analysis from Water

*A certified copy of the Method Validation report residing under the ASCI Study ID# 5030-003-09 is appended. The report contains a total of 27 pages which are numbered consecutively from p. 1 - p. 27 and also is numbered as "Page ___ of ___". These 27 pages are a part of the total number of pages included in this report.

STUDY TITLE

ISOOCTYL ACRYLATE: METHOD VALIDATION FOR ANALYSIS FROM WATER

AUTHORS

Minren Xu and Dinesh Vaishnav

STUDY COMPLETED

May 28, 1992

TESTING FACILITY

AScI Corporation
AScI-Duluth Environmental Testing Division
112 East Second Street
Duluth, MN 55805

Tel. No. (218) 722-4040

STUDY IDENTIFICATION NUMBERS

AScI Study ID# 5030-003-01

3M Company Study ID# J2774

CERTIFIED COPY

Signature: Date: 5/27/92

Sponsor: 3M Company
Sponsor Study ID# J2774

CERTIFICATION OF GOOD LABORATORY PRACTICE COMPLIANCE

To the best of my knowledge, this study was conducted in accordance with OECD Good Laboratory Practice Standards (OECD Council Decision C(81)30, Annex 2: OECD Principles of Good Laboratory Practice 1981).

Study Director:_____ Date:_____
Minren Xu
ASci Corporation/ASci-Duluth
Environmental Testing Division

Based on the signatures of the Study Director and the Quality Assurance Auditor, this study, to the best of our knowledge, was conducted in accordance with OECD Good Laboratory Practice Standards (OECD Council Decision C(81)30, Annex 2: OECD Principles of Good Laboratory Practice 1981).

Sponsor:_____ Date:_____

Submitter:_____ Date:_____

STATEMENT OF QUALITY ASSURANCE

The study data were reviewed by the ASCI-Duluth Environmental Testing Division Quality Assurance Unit to assure that standard operating procedures and guidelines used to conduct this study were followed, and this report is an accurate reflection of the raw data. The types of audits performed are listed in the following table.

Type of Audit for ASCI Study ID# 5030-003-01	Audit Date	Date Reported to Study Director and Management
Study Plan	12-17-1991	12-17-1991
In-Life Phase	12-19-1991	12-19-1991
Raw Data and Draft Report	01-09-1992	01-09-1992
Final Report	05-28-1992	05-28-1992

_____ Date: _____

Alan Mozol
Acting Manager, Quality Assurance Unit

TABLE OF CONTENTS

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STUDY SUMMARY TABLE

Study Title	Isooctyl Acrylate: Method Validation for Analysis from Water
Good Laboratory Practice Standards	As promulgated under the OECD Council Decision C(81)30, Annex 2: OECD Principles of Good Laboratory Practice (OECD 1981).
Sponsor	Rich Purdy, 3M Environmental Laboratory, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55106; Tel No. (612) 778-5379.
Sponsor's Representative	Susan A. Beach, 3M Environmental Laboratory, Building 2-3E-09, 935 Bush Avenue, St. Paul, MN 55106; Tel No. (612) 778-7452.
Testing Facility	ASCI Corporation/ASCI-Duluth Environmental Testing Division, 112 East Second Street, Duluth, MN 55805; Tel. No. (218) 722-4040.
Study Director	Minren Xu
Acting QAU Manager	Alan Mozol
Testing Facility Director	Donald Mount
Study Initiation Date	December 17, 1991
Test Dates	December 17-19, 1991
Test Substance	Isooctyl acrylate (CAS No. 29590-42-9[ ]Lot 3290), 99.75% acrylate (as determined by Sponsor [ ]liquid.

Test Description	Calibration Curves: (1) Standard solutions of various test substance concentrations and reagent (acetone) blank were prepared in acetone, (2) all solutions and reagent blank were analyzed twice by GC/MS, and (3) data were used to calculate regression equations, analytical method detection limits and other statistics.
Test Description (continued)	Spike Solutions and Recoveries: (1) Three replicates of test substance low and high spike solutions, and method blank (deionized water) were prepared using deionized water, (2) spike solutions and method blank were extracted using solid/liquid extraction technique, and extracts analyzed by GC/MS, and (4) data were used to calculate test substance recoveries from spike solutions.

Test Results	Percentage relative standard deviation (% RSD): First calibration curve -- 0.81% Second calibration curve -- 1.93% Correlation coefficient (r): First calibration curve -- 1.000 Second calibration curve -- 0.999 Method detection limit (MDL): With first calibration curve -- 0.04 mg/L With second calibration curve -- 0.04 mg/L Mean percentage recovery (R) from low spike solution (0.123 mg/L test substance): 85.91% Mean percentage recovery (R) from high spike solution (8.8 mg/L test substance): 103.48% Combined mean percentage (R) recovery from low and high spike solutions: 94.70%
Location of Raw Data and Final Report	ASci Corporation/ASci-Duluth Environmental Testing Division, 112 East Second Street, Duluth, MN 55805; Tel. No. (218) 722-4040.

1.0 INTRODUCTION

The test substance, isooctyl acrylate, is an ester made from primarily isooctanol and acrylic acid. According to OECD recommendations for new chemical substances (OECD Council Decision, 12th May, 1981; C(81)30), (1) the test substance physical-chemical properties and toxicities to various aquatic organisms need to be determined, and (2) chemical effects must be reported on the basis of measured chemical concentration. For the latter, there was a need to validate an analytical method so that test substance concentration can be determined from matrices employed in various tests. The analytical method was provided by the Sponsor.

The objectives of the present study were: (1) to develop an acceptable calibration curve, (2) to calculate detection limit of the analytical method, and (3) to determine test substance recoveries from spike solutions prepared using deionized water.

2.0 TEST METHODS

2.1 Formulas and Definitions. The formulas and definitions used in this study were:

(1) Test Substance Mean Percentage Recovery (R)

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R_i = (Measured concentration/Target concentration) X 100

The mean R was calculated using individual R_i values which fell within R ± 3SD range. If, the mean R was not between 80% and 120%, all measured concentrations were corrected accordingly.

(2) Method Detection Limit (MDL)

MDL = 3 X background signal in reagent blank

(3) Relative Standard Deviation of Calibration Curve (% RSD)

% RSD = (Standard deviation of slope/slope) X 100

(4) The sample response was corrected for the response of the method blank, if interference from the method blank was expected to have any effect on the sample response.

2.2 Test Substance. The test substance, isooctyl acrylate, (CAS No. 29590-42-9[ ]Lot 3290) was received at ASci on October 3, 1991 in one amber glass bottle placed in a sealed metal container. The test substance was stored at room temperature as received. According to a material safety data sheet and a written communication provided by the Sponsor, (1) the test substance was a clear, colorless, mobile liquid with acrylate odor, (2) the test substance concentration in deionized water can be analyzed by a GC method, (3) the test substance was 99.7% acrylate as determined by Sponsor[ ]and (4) the test substance had 1 mm Hg vapor

pressure at 50°C. The Sponsor also had information that based on the chemical structure, there would be essentially no dissociation or pH-dependent hydrolysis of the test substance at environmental pH levels.

2.3 Apparatus and Reagents. The apparatus and reagents used were:

(1) HP model 5890 gas chromatograph with 30 m 0.32 DB-5 (J & W Scientific) capillary column;

(2) HP model 5970 mass spectrometer;

(3) Pesticide grade methylene chloride and other solvents;

(4) Deionized water; and

(5) Extraction apparatus.

2.4 GC/MS Analysis. The analytical conditions were:

(1) Carrier gas: Helium at a total inlet purge flow of 40 ml/minute and a septum purge flow of 1 ml/minute with splitless injection mode;

(2) Temperature program: Isothermal at 70°C for 2 minutes then 8°C per minute to 200°C;

(3) Ionization source: Electron impact with a scan range of 20-500 mμ; and

(4) Detection method: Total ion chromatograph.

Before analysis, mass spectrometer was tuned using autotune program. A GC column performance test was conducted using column check sample (HP Sample A) to meet the criteria recommended by the manufacturer. A post GC/MS performance test was carried out by running a column check sample (HP Sample A) to ensure the stability of the instrument during the analytical test.

2.5 Calibration Curve. Two test substance stock solutions were prepared in acetone in 10-ml volumetric flasks. The first solution contained 1,760 mg/L test substance and the second solution contained 880 mg/L test substance. Subsequently, four standard solutions were prepared by adding appropriate volumes of the second stock solution to 10-ml volumetric flasks and diluting to volume with acetone. A reagent blank was prepared using acetone.

Each stock and standard solution, and reagent blank were analyzed twice by GC/MS. The instrument responses, except of reagent blank, from 8.95 to 12.958 minutes were integrated using a group integration method, and correlated with the test substance nominal concentration. The relative standard deviations of calibration curves (% RSD) and method detection limits (MDL) were then calculated.

2.6 Spike Solutions. Three replicates of a low level spike solution were prepared by adding 7 µl of test substance second stock solution (880 mg/L) to 50 ml of deionized water. This produced a target spike concentration of 0.123 mg/L test substance. Similarly, three replicates of a high level spike solution were prepared by adding 5 ml of test substance second stock solution (880 mg/L) to 500 ml of deionized water. This produced a target spike concentration of 8.8 mg/L test substance. A method blank was prepared using 500 ml of deionized water.

2.7 Test substance Extraction and Analysis. Both spike solutions and method blank were first extracted, using solid/liquid extraction procedure, and extracts analyzed by GC/MS. The extraction procedure was:

(1) Placed a 25-mm (with 50 ml sample) or 47-mm diameter (With > 50 ml sample) Empore™ extraction disk (J.T. Baker, Inc.) between a filter base and reservoir;

(2) Pre-washed the disk with 10 ml of methylene chloride (elution solvent);

(3) Applied vacuum to draw the solvent through the disk;

(4) Added 10 ml of methanol, applied vacuum and left a meniscus of methanol just above the top of the disk *(NOTES: RELEASED VACUUM BEFORE THE DISK WAS DRY. DID NOT*

ALLOW DISK TO DRY AT ANY TIME BEFORE SAMPLE FILTRATION WAS COMPLETED);

(5) Added 20 ml of deionized water to the reservoir, applied vacuum and left a meniscus of water just above the top of the disk;

(6) Added 5 ml methanol per liter of sample and mixed well;

(7) Poured sample into the reservoir and applied vacuum. The minimum filtration time was 10 minutes/L of sample;

(8) After the sample was processed, drew air through disk for 15 minutes;

(9) Placed the tip of the filter base into a test tube inside the filtration flask;

(10) Rinsed the volumetric flask with 2.5 ml (with 50 ml sample) or 4-5 ml (with ~ 50 sample) methylene chloride and added the solvent to the reservoir;

(11) Drew half the solvent through the disk and let stand for approximately 1 minute. Drew the remainder through the disk;

(12) Repeated Steps 10 and 11 three times;

(13) Collected a measured volume of methylene chloride extract; and

(14) Processed the method blank in the same way (Steps 1 to 13) as the sample.

For low spike solutions, extracts were first concentrated under a gentle stream of nitrogen gas and the volumes of concentrated extracts measured. The extracts of both low and high spike solutions were then transferred to analytical vials and analyzed for the test substance concentrations using the GC/MS instrument. The instrument was operated as per manufacturer's recommendation.

2.8 Test Substance Recovery. The instrument responses between 8.95 and 12.958 minutes were integrated using a group integration method, and fitted to the first calibration curve to determine test substance concentrations. These data were then used to calculate the test substance percentage recoveries from spike solutions.

2.9 Test Substance Analysis During Various Tests. Several physical/chemical and toxicity tests were performed separately with this test substance. In analyzing the test substance concentrations in aqueous samples from these tests, the following procedure was used:

(1) At each test initiation, developed an acceptable new calibration curve with a relative standard deviation (% RSD) within 10%;

(2) Each day when test substance concentrations in aqueous samples from a particular test were analyzed, re-validated the previous calibration curve (from Step 1) using at least two standard solutions, or developed a new acceptable calibration curve with a relative standard deviation (% RSD) within 10%. In case of re-validation, the previous calibration curve was considered valid and the same regression equation (From Step 1) was used, if the measured and nominal concentrations of standard solutions did not differ by more than 10%;

(3) Each time when test substance concentrations in aqueous samples from a particular test were analyzed, standard (deionized water) and test (e.g. well water, algal medium etc.) matrices blanks, and spiked standard and test matrices were prepared. The test substance spike concentration was close to the lowest nominal concentration used in a particular test. Generally, the spike concentrations were similar to the low spike concentration (0.123 mg/L) used in this method validation study;

(4) Analyzed both standard and test matrices and calculated percentage spike recoveries;

(5) Accepted spike recoveries if they were within the same range (85.91 ± 22.859%) as low spike recovery established from this method validation study;

(6) Each time when test substance concentrations in aqueous samples from a particular test were analyzed, corrected (to 100%) test substance concentrations in aqueous samples for the percentage matrix spike recovery for that time.

2.10 Data Analysis. All data were analyzed using Minitab® statistical software (Minitab, Inc. 1988), MS ChemStation software (HP 1990) which interfaced the GC/MS instrument, and a scientific calculator.

3.0 RESULTS

Six test substance solutions, including two stock and four standard solutions (Table 1), were used to prepare two calibration curves. The use of a broad range of solution concentrations was important because the test substance concentrations in biological tests are expected to range from approximately 0.1 mg/L to the test substance water solubility concentration (12.44 mg/L).

The samples from physical/chemical and biological tests will be extracted and test substance concentrations eluted in approximately 15 ml of solvent (actual extract volume will be measured). Accordingly, one solution (standard solution 1) used for the two calibration curves had a test substance concentration approximately 3 fold greater than the method detection limit (MDL) of 0.04 mg/L (Table 1). All other solutions, except the first stock solution, were below and near the test substance solubility (12.44 mg/L) in deionized water (Table 1). The test substance concentration in the first stock solution was approximately twice the solubility concentration.

The GC/MS responses in two calibration curves are listed in Table 2. Correlations of GC/MS response (ordinate) and test substance nominal concentration (abscissa) had correlation coefficients (r) of 1.000 and 0.999 for the first and second calibration curves, respectively (Table 3). The slopes from both curves differed by approximately 0.32%, and relative standard deviations (% RSD) of slopes were 0.81% and 1.93% for the first and second calibration curves, respectively (Table 3). The detection limit of 0.04 mg/L test substance was the same as calculated for both calibration curves (Table 3).

The low spike concentration was 0.123 mg/L test substance and high level spike concentration was 8.8 mg/L test substance (Table 4). These concentrations were within the range of test substance concentrations to be used in biological and physical/chemical tests. The volumes of spike solutions (50 ml and 500 ml) used were comparable to the volumes that may be analyzed from physical/ chemical and biological studies. The test substance recoveries for the low spike solution ranged between 70.73% and 112.20% with a mean of 85.91 ± 22.859%, and for the high spike solution between 97.50% and 111.36% with a mean of 103.48 ± 7.121%. (Table 4). The combined mean recovery for low and high spike solutions was 94.70 ± 17.943% (Table 4).

The test substance concentration in the method blank was below the method detection limit of 0.04 mg/L isooctyl acrylate.

From the quality assurance standpoint, this test is acceptable because it complies with the acceptance criteria (Table 5).

4.0 CONCLUSIONS

The GC/MS response and test substance, isooctyl acrylate, concentrations between 8.8 and 1,760 mg/L were in linear

correlation. The test substance combined mean recovery (94.70%) from low and high spike solutions suggested that extraction and analytical procedures should be adequate for use with other aqueous samples.

5.0 DEVIATIONS FROM APPROVED ASCI STUDY PLAN

The deviations which occurred while conducting this study were:

(1) HP model 5890 gas chromatograph and HP model 5970 mass spectrometer were used instead of HP model 5970 gas chromatograph and HP model 5890 mass spectrometer.

(2) In GC/MS analysis, total inlet purge flow of helium gas was at 40 ml/minute and a septum purge flow was at 1 ml/minute, instead of helium at 5.5 ml/min and a septum purge flow of 5.8 ml/minute.

(3) In GC/MS analysis, temperature program used was 70°C for 2 minutes and then 8°C/minute to 200°C, instead of 70°C for 2 minutes, and then 8°C/minute to 220°C and holding at 220°C for 2 minutes, or as appropriate. This was because after 180°C nothing eluted from the GC column.

To the best of our current scientific knowledge and understanding, this deviation should have no effect on the results presented in this report.

6.0 REPORT SIGNATURE

Study Director:_____Date:_____
Minren Xu
ASci Corporation/ASci-Duluth
Environmental Testing Division

7.0 REFERENCES

Hewlett Packard (HP). 1990. HP 59940A MS ChemStation (HP-UX series) Handbook.

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Organization for Economic Cooperation and Development (OECD). 1981. OECD Guidelines for Testing of Chemicals. OECD Publication Information Center, Washington, DC.

8.0 PERSONNEL INVOLVED IN STUDY AND THEIR RESPONSIBILITIES

Personnel	Responsibility
Minren Xu	Study Director
Connie Coleson	Glassware preparation
Billie Samson	Laboratory assistance
Dinesh Vaishnav	Report preparation
Alan Mozol	QAU
Nancy Jordan	Archivist

Table 1. Isooctyl acrylate (test substance): Solutions for two calibration curves

Test substance solution	Dilution	Test substance nominal concn (mg/L)
Reagent blank	0.0 µl test substance in 10 ml acetone (final volume)	0.0
First stock solution	20 µl test substance in 10 ml acetone (final volume)	1,760
Second stock solution (SS)	25 µl test substance in 25 ml acetone (final volume)	880
Standard solution 1	100 µl SS in 10 ml acetone (final volume)	8.8
Standard solution 2	500 µl SS in 10 ml acetone (final volume)	44
Standard solution 3	1,000 µl SS in 10 ml acetone (final volume)	88
Standard solution 4	5 ml SS in 10 ml acetone (final volume)	440

Table 2. Isooctyl acrylate (test substance): GC/MS responses in two calibration curves

Test substance nominal concn (ng/L)	GC/MS response in first calibration curve	GC/MS response in second calibration curve
Reagent blank	19,622	19,622
1,760	2,719,632,005	2,729,584,720
880	1,390,089,059	1,258,512,351
8.8	22,481,557	10,280,168
44	62,891,391	52,827,478
88	128,917,851	113,808,095
440	658,002,779	622,643,636

Table 3. Isooctyl acrylate (test substance): Statistical analysis of two calibration curves[a]

Parameter	First calibration curve	Second calibration curve
Regression equation	-1.76e+06 + 1.55e+06 (x)[b]	-2.48e+07 + 1.54e+06(x)[b]
Slope ± SD	1551104 ± 12498[c]	1546151 ± 29918[c]
Relative standard deviation (% RSD)[d]	0.81%	1.93%
Correlation coefficient (r)	1.000	0.999
Method detection limit (MDL)[e]	0.04 mg/L	0.04 mg/L

[a]GC/MS response and isooctyl acrylate (test substance) concentration (milligrams per liter) were plotted on ordinate and abscissa, respectively.

[b]Equation was generated using MS ChemStation software (HP 1990).

[c]Slope and SD were calculated using Minitab® statistical software (Minitab, Inc. 1988), as HP-UX software did not calculate SD.

[d]Percentage RSD = (Standard deviation of slope/slope) X 100.

[e]MDL = 3 X response in reagent blank (= 19,622; Table 2)/slope.

Table 4. Isooctyl acrylate (test substance): Recoveries from spiked deionized water

Type of solution	Rep	Test substance target concn (mg/L)	Test substance measured concn (mg/L)[a]	% Recovery (R_i)[b]	Mean ± SD% recovery ($\bar{R}$)[c]
Method blank	1	0.0	<0.04[d]	-	-
Low spike	1	0.123	0.092	74.80	
	2	0.123	0.138	112.20	85.91 ± 22.859
	3	0.123	0.087	70.73	
High spike	1	8.8	8.58	97.50	
	2	8.6	8.94	101.59	103.48 ± 7.121
	3	8.6	9.80	111.36	
Combined recovery from low spikes + high spikes					94.70 ± 17.943

[a]Determined using first calibration curve (Table 3).

[b]R_i = (Measured concentration/Target concentration) X 100.

[c]Mean R was calculated using R values which fell within $\bar{R}$ ± 3SD range.

[d]Method detection limit (MDL) was 0.04 mg/L isooctyl acrylate.

Table 5. Isooctyl acrylate (test substance): QA criteria and test acceptability

QA criterion	Results
Relative standard deviation of calibration curve (% RSD) must be within 10% Post run standard response must be within 10% of the same standard analyzed at the beginning of the test	% RSD of first calibration curve was 0.61% and of second calibration curve was 1.93% Responses from all peaks from post run standard differed by 5.95% compared to the beginning of the test

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Signature: Dinunb Date: 5/27/92